

What Maintains the Metabolic Cost at Peak Anaerobic Test in Elite Young and Master Male Cyclists?

Running Head: Anaerobic and VO₂ in Young and Masters.

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ABSTRACT

Purpose: The present study investigated the effect of aging on the oxygen utilization in healthy well-trained elite cyclists males young and masters at peak Wingate Anaerobic Test. **Methods:** Twenty young 21.21 ± 1 and twenty masters 57.6 ± 3.4 years were studied by Echocardiography performed with the subject sitting on a bicycle ergometry. Arterial oxygen was defined from echocardiographic measurements and venous oxygen content. **Results:** At peak Wingate Anaerobic Test, in young compare to masters, cardiac output (18.2 ± 0.6 and 17.1 ± 0.4 L•min⁻¹ respectively), and stroke volume (84.0 ± 5.0 and 78.2 ± 6.1 mL respectively) were significantly ($P < 0.05$) higher, (a-v)O₂ was significantly ($P < 0.05$) lower (122.5 ± 2.9 and 126.1 ± 3.1 mL O₂ respectively) while %VO₂ out of total energy utilized did not differ between groups. **Conclusions:** This study suggests that elite cyclist athlete young and masters respond to the Wingate Anaerobic Test by reducing their left ventricular systolic function in the face of increased afterload. The lack of differences at peak exercise in the oxygen utilization may be attributed to differences in energy metabolism, namely higher oxygen extraction in the masters, compensating for the lower oxygen delivery.

Key words: Strenuous exercise, Fick equation, oxygen extraction.

INTRODUCTION

During heavy exercise that engenders a significant lactic acidosis, oxygen uptake does not achieve steady state but continues to rise until exercise is terminated, or exhaustion ensues (Reis JF, Alves FB, Bruno PM, Vleck V, Millet GP. Effects of aerobic fitness on oxygen uptake kinetics in heavy intensity swimming. Eur J Appl Physiol. 2012; 112:1689-1697; Sousa A, Rodríguez FA, Machado L, Vilas-Boas JP1, Fernandes RJ. Exercise modality effect on oxygen uptake off-transient kinetics at maximal oxygen uptake intensity. Exp Physiol. 2015; 100:719-729). The nature of the contractile and metabolic properties of skeletal muscle suggests that the speed of shortening will influence energy turnover during contractions. Indeed, it has been observed in vitro using isolated muscles and single muscle fibers that energy varies with shortening velocity (He Z-H, Bottinelli R, Pellegrino MA, Ferenczi MA, Rrgigiani C. ATP consumption and efficiency of human single muscle fibres with different myosin isoform composition. Biophysical Journal 2000; 79:945-961; Widrick JJ, Stelzer JE, Shoepe TC, Garner DP. Functional properties of human muscle fibers after short-term resistance exercise training. Am J Physiol Regul Integr Comp Physiol. 2002;283: R408-416).

Metabolic and left ventricular responses during incremental exercise differ between men and women despite a similar fitness (Ben-Sira, Sagiv, M. The effect of gender on left ventricular systolic function at Peak Wingate Anaerobic Test. *Eur. J. Appl. Physiol.*1977; 75:549-553; Scott JM, Esch BT, Haykowsky MJ, Isserow S, Koehle MS, Hughes BG, Zbogor D, Bredin SS, McKenzie DC, Warburton DE. Sex differences in left ventricular function and beta-receptor responsiveness following prolonged strenuous exercise. *Appl Physiol* (1985). 2007; 102:681-687). In healthy subjects, it has been found that during aerobic exercises oxygen uptake of men exceeds that of women due to gender related variance in heart size (Ben-Sira, Sagiv, M. The effect of gender on left ventricular systolic function at Peak Wingate Anaerobic Test. *Eur. J. Appl. Physiol.*1977; 75:549-553; Sagiv M.; Amir O; Ben-Sira D, Amir R. Oxygen delivery of men and women at peak wingate test. *Brazilian Journal of Biomotricity.* 2007;1: 65-75).

During heavy exercise that engenders a significant lactic acidosis, oxygen uptake does not achieve steady state but continues to rise until exercise is terminated, or exhaustion ensues (Reis JF, Alves FB, Bruno PM, Vleck V, Millet GP. Effects of aerobic fitness on oxygen uptake kinetics in heavy intensity swimming. *Eur J Appl Physiol.* 2012; 112:1689-1697; Sousa A, Rodríguez FA, Machado L, Vilas-Boas JP, Fernandes RJ. Exercise modality effect on oxygen uptake off-transient kinetics at maximal oxygen uptake intensity. *Exp Physiol.* 2015; 100:719-729). Anaerobic all-out exercise such as the Wingate Anaerobic Test, exposes the subjects to a very high degree of sudden strenuous exercise relying primarily on the glycolysis pathway (Nummela A, Hamalainen I, Rusko H. Comparison of maximal anaerobic running tests on a treadmill and track. *J Spoers Sci* 2007; 25:87-96; Sahlin K. Muscle energetics during explosive activities and potential effects of nutrition and training. *Sports Med* 2014;44 (suppl 2): S167-173). Accordingly, performance time can be limited by lactic acid levels in the blood and active muscles (Vale RG, de Oliveira RD, Pernambuco CS, de Meneses YP, Novaes Jda S, de Andrade Ade F. Effects of muscle strength and aerobic training on basal serum levels of IGF-1 and cortisol in elderly women. *Arch Gerontol Geriatr* 2009;49:343-347; Girard O, Mendez-Villanueva A, Bishop D. Repeated-sprint ability - part I: factors contributing to fatigue. *Sports Med* 2011; 41:673-694). With aging two main factors influence the physical performance: a) the decrease in cardiopulmonary function and b) a progressive decline in muscle mass and strength that occurs with aging (Gregorich ZR, Peng Y, Cai W, Jin Y, Wei L, Chen AJ, McKiernan SH, Aiken JM, Moss RL, Diffie GM, Ge Y. Top-down targeted proteomics reveals decrease in myosin regulatory light-chain phosphorylation that contributes to sarcopenic muscle dysfunction. *J Proteome Res* 2016; 15:2706-2716). Consequently, the maximal oxygen uptake and power ability decrease, irrespective of lifestyle because of genetic factors, thereby severely limiting body function (Baumann CW, Kwak D, Liu HM, Thompson LV. Age-induced oxidative stress: how does it influence skeletal muscle quantity and quality? *J Appl Physiol* 1985, 2016;121:1047-1052). However, structured endurance training after 40 years of age is powerful enough to induce beneficial cardiovascular adaptations in later life (Matelot D, Schnell F, Kervio G, Ridard C, Thillaye du Boullay N, Wilson M, Carre F. Cardiovascular benefits of endurance training in seniors: 40 is not too late to start. *Int J Sports Med* 2016;3 7:625-632).

Although data are available on metabolic cost during maximal anaerobic dynamic exercise testing (Hill DW, Smith JC. Gender difference in anaerobic capacity: role of aerobic contribution. *Br J Sports Med.* 1993; 27: 45-48; Sagiv M, Amir O, Ben-Sira D, Amir R, Oxygen delivery of men and women at peak Wingate Test. *Brazilian Journal of Biomotricity.* 2007; 3:65-75), little data are available on the physiological variables that maintain the source of metabolic costs with

aging. Therefore, the purpose of the present study was to detect the effect of aging on exercise oxygen utilization in elite young men cyclist and master men cyclists during the 30-s all-out Wingate Anaerobic Test.

METHODS

Subjects: Twenty elite young men cyclists 21.2 ± 1.1 and twenty masters men cyclists 57.63 ± 4.4 years volunteered to participate in this study. All subjects were judged free of cardiac problems from their clinical history, absence of major risk factors and normal responses during graded exercise stress test up to their maximal oxygen uptake. A written informed consent approved by the Clinical Science Center Committee on Human Subjects, was obtained from each subject after being fully informed of the details and possible discomforts associated with the experimental protocol.

Procedure and measurements: Each subject reported to the laboratory 2 times. Sessions were spaced at least 48 hrs apart and on average at intervals of no more than 1 week. The first session was devoted to accustoming the subjects to the study procedures and to the general scope of the study. Following the explanations, subjects underwent a graded maximal bicycle exercise test on a mechanical weight-adjusted Monark cycle-ergometer (Model 818). End point for the maximal test was when further increment in workload did not increase oxygen consumption or when the subject could not keep up with the rate of work according to the guidelines of the American College of Sports Medicine (American College of Sports Medicine. ACSM's guidelines for exercise testing and prescription. Philadelphia, PA: Lippincott Williams & Wilkins; 2000. pp. 145-147 and 165-199).

Oxygen uptake was determined breath by breath utilizing the Medical Graphics (St. Paul, MN) metabolic cart. The metabolic cart was calibrated before each test with known primary standard quality gases. A 12-lead electrocardiogram and heart rate were continuously monitored at rest, during exercise and recovery. Five-second recordings were obtained at rest and at peak exercise. Following warm-up, subjects cranked against an initial work rate of 150 watts which was increased by 25 watts every minute until the subject could no longer continue at the predetermined pace. Blood pressure was taken using a standard sphygmomanometer cuff and mercury manometer mounted at eye level, in the sitting position at rest and at peak exercise.

During the 2nd session subjects were asked to perform the 30 second all out Wingate Anaerobic Test (Bar-Or O, Dotan R, Inbar O, Rotstein A, Karlsson J, Tesch P. Anaerobic capacity and muscle fiber type distribution in non-athletes and athletes. *Int J. Sports Med.* 1980; 1:82-85). The end point for the Wingate anaerobic test was at the end of the 30 seconds. Following warm-up, subjects were asked to perform the 30 second all-out Wingate Anaerobic Test, utilizing a weight-adjusted Monark cycle-ergometer (Model 864). Subjects were seated on the ergometer with their feet fastened to the pedals by means of racing-type toe-clips, and seat height was adjusted. In addition, subjects' backs were strapped to the wall, in order to minimize upper body movement and to facilitate auscultation of blood pressure and echocardiographic measurements at peak exercise.

The anaerobic test consisted of 30 seconds supramaximal pedaling against a resistance determined relative to the subject's body mass at $80 \text{ gr} \cdot \text{kg}^{-1} \cdot \text{body weight}^{-1}$ (Smith JC, Hill DW.

Contribution of energy systems during a Wingate power test. *Br J Sports Med.* 1991; 25:196-199). At the command “start”, the subjects commenced cranking as fast as they could against the ergometer’s inertial resistance only. The full, predetermined resistance load was applied within 3-4 seconds once the inertial resistance had been overcome. Pedal revolution count started at that instant by means of an electro-mechanical counter and subjects maintained an all-out effort throughout the test. Strong verbal encouragement was given to ensure maximal effort.

During the 1st and 2nd testing sessions, Time of day for testing was kept consistent for all subjects to control for problems associated with diurnal variations.

A 25 μ fingertip blood sample was taken at rest and at peak anaerobic effort at the 3rd minute post exercise for the determination of lactic acid concentration. The sample was immediately transferred to a micro-tube containing 100 μ of 7% perchloric acid. The tubes were centrifuged after standing for at least 1 hour. Twenty microliter aliquots of the supernatant were subsequently used for lactic acid analysis on the Analox LM3 analyzer (Analox Instruments, England; Reagent Kit No. GMRD-071).

Echocardiographic data processing: 2-D, echocardiographic and M-mode images were performed at rest and at peak of the Wingate anaerobic test, utilizing Vingmed 725 Sonotron and Sony recorder equipped with 2 and 3 MHz transducers. The diameters of the aorta were determined by two-dimensionally directed M-mode. The left atrium was measured from the parasternal long-axis view. At rest, left ventricular end-diastolic and end-systolic diameters and intraventricular septum and left ventricular posterior wall thicknesses were measured from the parasternal long and short-axis views as well as from 4 and 5 chamber views, just below the mitral valve level, according to the recommendations of the American Society of Echocardiography (Shaw DJ, DeMaria A, Kisslo J, Weyman A. Recommendations regarding quantitation in M-mode echocardiography: Results of survey of echocardiographic measurements. *Circulation.* 1978; 58:1072-1083). At peak Wingate Anaerobic Test, due to the short time available for measurements, left ventricular volumes and ejection fraction were determined using Simpson’s rule from apical 4 chamber view.

All echocardiographic studies were performed with the subjects in the sitting position. The probe was handheld and directed to a marked point from which the resting data were obtained. The beam was directed to the aortic valve outflow tract in the 5-chamber view, or from the substernal approach for those subjects in whom adequate imaging of 5-chamber or parasternal long axis views was not obtained. To assess the objectivity of the echocardiographic readings, all recordings were evaluated by two independent experts. A high correlation ($r = 0.93$) was found for inter-observer reliability.

CALCULATIONS

At rest and during exercise variables were computed as follow:

Stroke volume was the difference of left ventricular end diastolic volume - end systolic volume. Cardiac output was determined as the product of heart rate and stroke volume.

Venous oxygen content ($\text{mLO}_2 \cdot \text{dL}^{-1}$) was calculated from the measured O_2 saturation, and Hb concentration multiplied by 1.34 mLO_2 .

Oxygen saturation and Hb concentration were measured independently in each blood sample (IOSM3 hemoximeter, Radiometer).

Arterial O_2 content was calculated utilizing the Fick equation, in which:

$$\text{VO}_2 = \text{HR} \times \text{SV} (\text{a-v}) \text{O}_2,$$

The arteriovenous O_2 difference (a-v) O_2 was calculated by subtracting venous O_2 content from the calculated arterial O_2 content, using VO_2 , cardiac output and venous O_2 content measured.

Left ventricular mass index was calculated according to Devereux and Reichek (17).

% VO_2 out of energy utilized was calculated by dividing watts achieved at peak exercise (converted to $\text{VO}_2 \text{L} \cdot \text{min}^{-1}$) by direct measured $\text{VO}_2 (\text{L} \cdot \text{min}^{-1})$.

RESULTS

All the subjects completed the 30-s study without any difficulties or clinical abnormalities. Descriptive data of the subject's physical characteristics are given in table 1. It reveals that at rest compared to masters, in the young group left ventricular mass index and end diastolic dimension were significantly ($p < 0.05$) higher. At peak maximal oxygen uptake test, the young group compared to the masters, achieved a significant ($p < 0.05$) higher maximal oxygen uptake, maximal heart rate and maximal cardiac output. Descriptive statistics are given in table 2, it revealed that at rest no significant differences were noted between the groups for all measured variables. At peak Wingate anaerobic test, in the young group, cardiac output lactic acid and workload were significantly ($P < 0.05$) higher than for the masters. Figures 1-3 reveal that before the Wingate anaerobic test bout, at rest, young compared to masters, have shown a significant ($P < 0.05$) higher stroke volume values while, (a-v) O_2 and VO_2 values did not differ significantly. At peak exercise, stroke volume in both groups was reduced significantly compared to resting values yet, young compared to masters, have shown a significantly ($P < 0.05$) higher values. At peak exercise, (a-v) O_2 achieved was significantly higher in the masters ($P < 0.05$) than that of the young. No significant differences between the groups were noted for VO_2 attained at peak exercise.

DISCUSSION

The present study tested the hypothesis that aging may influence the metabolic sources at peak anaerobic bout in healthy well-trained young and master men. Exercise oxygen uptake is maintained because of a balance between oxygen delivery and extraction.

The present study did not reveal significant differences between both groups in absolute oxygen uptake values at peak Wingate Anaerobic Test due to aging. However, one has to keep in mind the significant higher workload and peak power output in the young group. Oxygen uptake in masters was compensated for the very lower cardiac output and stroke volume by a significant increase in (a-v) VO_2 .

Both groups achieved similar absolute oxygen uptake values attained at peak exercise which accounts for 20% of the total energy expenditure in both groups. All energy systems are active at a given time, the primary energy source utilized depends on the intensity and duration.

The maximal (a-v) $\dot{V}O_2$ decreases from perhaps 140-150 mL $\cdot$ dL⁻¹ in a young adult to 120-130 mL $\cdot$ dL⁻¹ in elderly, thus, in untrained elderly (a-v) $\dot{V}O_2$ is limited (Shephard RJ. *Health and Aerobic Fitness*. Champaign, IL.: Human Kinetics Publishers, 1993.).

With the reduced cardiac delivery in the masters compared to the young once, the similar oxygen uptake reached at peak Wingate Anaerobic Test may have been due to different muscle fiber recruitment patterns or different efficiencies of fibers that were recruited during exercise. Previous findings indicated that in untrained normal population, young have larger muscle fibers size than aged subjects, however, training increase entirely muscle fiber hypertrophy in particular type II muscle fiber (Nilwik R, Snijders T, Leenders M, Groen BB, van Kranenburg J, Verdijk LB, van Loon LJ. The decline in skeletal muscle mass with aging is mainly attributed to a reduction in type II muscle fiber size. *Nilwik R. Exp Gerontol.* 2013; 48:492–498). These differences may influence metabolism and the adaptive response to exercise (19). In addition, it seems that the lower velocity of contraction in the masters compared to the young enabled the masters to utilized relatively slower twitch fibers and hence increasing the ability to extract oxygen. From in vitro studies, it has been observed that the efficiency of different fiber types is closely related to the velocity of contraction (HE Z.-H., BOTTINELLI R, PELLEGRINO MA, FERENCZI MA, REGGIANI C. ATP consumption and efficiency of human single muscle fibres with different myosin isoform composition. *Biophysical Journal* 2000; 79:945-961). At low contraction velocities as in the masters, the efficiency of slow twitch fibers is higher than for fast twitch fibers and the reverse is observed at high speeds (HE Z.-H., BOTTINELLI R, PELLEGRINO MA, FERENCZI MA, REGGIANI C. ATP consumption and efficiency of human single muscle fibres with different myosin isoform composition. *Biophysical Journal* 2000; 79:945-961).

Values for cardiac output during the Wingate anaerobic test, were low in both groups compared to what one would have expected during maximal dynamic exercise. The explanation for the lower cardiac output in young subjects and to the greater extend in masters during the Wingate Anaerobic Test might be related to the short duration of the bout, which in turn it did not bring about the appropriate adjustment of the circulation, due to the relatively high total peripheral resistance which was not as low as seen during aerobic exercise.

Ecocardiographic indices and blood pressure response in the present study were similar to those seen during isometric exercise, indicating that autoregulation is not involved while performing sudden strenuous all-out exercise. This assumed lack of auto-regulation mechanism may indicate that the reduced stroke volume in the young and masters during the Wingate anaerobic test was due to the relatively high-pressure overload opposing left ventricular ejection. Cardiac adaptations in elderly following aerobic exercise training improves stroke volume and left ventricular systolic function at peak aerobic exercise (Carrick-Ranson G, Hastings JL, Bhella PS, Fujimoto N, Shibata S, Palmer MD, Boyd K, Livingston S, Dijk E, Levine BD. The effect of lifelong exercise dose on cardiovascular function during exercise. *J Appl Physiol* 2014; 116:736-745).

Although in elderly subject systolic function is preserved, stroke volume reduction at peak exercise was significantly larger in the masters compared to the young due, to the lesser increased contractility attributed to functional changes with age in the myocardium (Sagiv M, Ben-Sira D, Sagiv M, Goldhammer E. Left ventricular function at peak all-out anaerobic exercise in older men. *Gerontology*. 2005; 51:122-125). This response is in agreement with previous reports which suggested that during strenuous exercise well trained healthy young adult subjects did not increase stroke volumes and sometimes stroke volume was reduced (Sagiv M, Ben-Sira D, Goldhammer E, Soudry M. Left ventricular contractility and function at peak aerobic and anaerobic exercises. *Med Sci. Sports Exerc*. 2000; 32:1197-1201), while elderly also increased their contractility but not to those values seen in the young subjects (Sagiv M, Ben-Sira D, Sagiv M, Goldhammer E. Left ventricular function at peak all-out anaerobic exercise in older men. *Gerontology*. 2005; 51:122-125).

CONCLUSIONS

This study suggests that elite cyclist athlete young and masters respond to the Wingate Anaerobic Test by reducing their left ventricular systolic function in the face of increased afterload. The lack of differences at peak exercise in oxygen utilization may be attributed to differences in energy metabolism, namely higher oxygen extraction in the masters, compensating for the lower oxygen delivery.

Legends for Illustrations

Figure 1: Stroke volume responses for young and masters at rest and peak anaerobic exercise.

Figure 2: oxygen uptake for women and men at rest and peak anaerobic exercise.

Figure 3: Oxygen extraction (a-v) O₂ responses for women and men at rest and peak anaerobic exercise.

Table 1: Physical characteristics of the subjects (mean $\pm$ SD).

VARIABLE	MEN	MASTERS	SIG
Weight (kg)	66.2 $\pm$ 3.7	64.4 $\pm$ 4.1	
Height (cm)	181.5 $\pm$ 4.3	178.2 $\pm$ 5.0	
Age (Years)	21.2 $\pm$ 1.0	57.6 $\pm$ 3.4	P < 0.01
LV mass index (g•m ²)	96.3 $\pm$ 5.9	97.8 $\pm$ 4.2	
Lean body mass (kg)	58.9 $\pm$ 3.0	56.7 $\pm$ 4.9	
Body fat (%)	10.8 $\pm$ 2.5	11.7 $\pm$ 3.0	
EDD (cm)	4.9 $\pm$ 0.3	4.7 $\pm$ 0.3	P < 0.05
ESD (cm)	3.1 $\pm$ 0.2	3.1 $\pm$ 0.3	
EF (%)	62.0 $\pm$ 5.0	58 $\pm$ 6.1	
VO ₂ max. (mL•kg ⁻¹ •min ⁻¹)	63.1 $\pm$ 6.3	54.4 $\pm$ 5.1	P < 0.04
Qmax. (L•min ⁻¹)	29.7 $\pm$ 1.2	25.6 $\pm$ 1.3	P < 0.05
HRmax. (beats•min ⁻¹)	213 $\pm$ 4.6	202 $\pm$ 5.3	P < 0.05
Max bout duration (min)	6.4 $\pm$ 0.4	6.3 $\pm$ 0.6	

LV = Left ventricular; EDD = End diastolic dimension; ESD = End systolic dimension; EF = Ejection fraction; VO₂ = Maximal oxygen uptake; Q = Cardiac output; HR = Heart rate.

Table 2: Hemodynamic responses and left ventricular function at rest and during Wingate Anaerobic Test in young and masters (mean±SD).

Variable	Rest		Exercise		Sig
	Men	Masters	Men	Masters	
HR (beats•min ⁻¹)	54.0±3.0	60.0±6.0	180.0±7.7	174.0±5.9	
Q (L•min ⁻¹)	5.6±0.3	5.0±0.5	18.2±0.6	17.1±0.4	a
LA (mmol•L ⁻¹)	1.4±0.2	1.3±0.4	12.6±0.7	11.5±0.8	a
SBP (mm Hg)	106.0±8.8	103.0±6.4	191.1±10.0	188±11.1	
DBP (mm Hg)	69.0±6.3	66.0±5.6	99.7±5.9	92.8±5.7	a
Load (Watts)	-----		390.5±24.1	358.0±14.6	a
%VO ₂ utilized	-----		20.0±0.4	20.0±0.4	

a = Significant difference between groups during exercise (P <0.05).

Note: Significant difference from rest to exercise were seen in both groups in all measured variables.



